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14. ABSTRACT

Jet propulsion fuel 8 (JP8) has recently been recognized by the Department of Defense as the single larges chemical exposure for its personnel. the primary aim of the project (OJENES, Occupational JP8 Exposure Neuroepidemiology study) is to conduct an epidemiological field study to examine the relationship between JP8 fuel exposure and adverse neurological health in military personnel. The research objectives include 1) determination of the individual service member's level of exposure ot JP8 components measured by specific biomarkers of exposure, and 2) examination of whether exposure to JP8 over a work week is significantly associated with hypothesized neurobehavioral and neurophysiologic performance outcomes. the projec thas two phases: Tier I involves onsite exposure assessment to fully characterize JP98 exposure parameters in the military occupational filed setting required for the planned field study; Tier II is the full0scale neuroepidemiology field study to examine predicted dose-collection and subsequent sample analysies fo the Tier I & II phases have been completed; Currently, nine manuscripts (from Tier I and II) are in press or in preparation.

15. SUBJECT TERMS Jet fuel, JP-8, neurobehavior, neurological health, exposure assessment

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Table of Contents

Introduction

Jet propulsion fuel 8 (JP8) has recently been recognized by the Department of Defense (DoD) as the single largest chemical exposure for its personnel. The primary aim of the project is to conduct an epidemiological field study to examine the relationship between JP-8 fuel exposure and neurological functioning in military personnel. The research objectives include 1) determination of the individual service member's level of exposure to JP-8 components while carrying out his/her job tasks, as measured by specified biomarkers of exposure, and 2) examination of whether acute, or cumulative exposure to JP-8 over a work week is significantly associated with hypothesized neurobehavioral and neurophysiologic performance outcomes. The project has two phases: Tier I is to conduct onsite exposure assessment techniques to fully characterize JP-8 exposure parameters in the military occupational field setting required for the planned field study; Tier II is the conduct of the full-scale neuroepidemiology field study to examine predicted dose-response relationships. The field study is being carried out with military (Air Force) personnel.

Body

Due to administrative delays that occurred at the beginning of this project, a request to modify the timeline of the study Statement of Work (SOW) was submitted in March 2007. Thus, the current, approved SOW (Table 1) incorporates those modifications and reflects the timeline of required tasks.

Table 1. Modified SOW, approved June, 2007

Year 1	**Months 1-12**	***Task 1***	-Obtain all required administrative approvals.
		Task 2	-Conduct planning steps, which include field site exposure measurements and samples analyzed.
		Task 3	-Convene Working Groups.
Year 2	**Months 13-24**	***Task 4***	-Conduct Tier I phase.
		Task 5	-Carry out analyses of environmental/biological samples from Tier I phase.
		Task 6	-Perform data management tasks to integrate multiple data sources for data analyses.
		Task 7	-Convene Workshop.
		Task 8	-Initiate Tier II phase.
Year 3	**Months 25-32**	***Task 9***	-Complete analyses of environmental and biological samples (Tier II).
		Task 10	-Complete data analyses of exposure-outcome hypothesis relationships (Tier II).
		Task 11	-Prepare Final Report and manuscript(s).

The project was funded November 1, 2005. The progress made during the first 8 months of the project was reported in the 2006 Annual Report. Specifically, ***Task 1***, obtaining the required Army and Air Force administrative approvals, was completed, and progress on ***Tasks 2 & 3*** described. Progress made during months 9-20 of the project was reported in the 2007 Annual Report. Specifically, ***Tasks 2-5*** were completed; ***Task 6*** was in progress. Progress made on tasks outlined in the modified SOW for months 21 through 32 was reported in the 2008 Annual Report. Specifically, ***Tasks 6-8*** were completed; ***Tasks 9-11*** were in progress. Progress made on ***Tasks 9-11*** was reported in the 2009 Annual Report. Specifically, ***Task 9*** was completed; ***Tasks 10-11*** were in progress.

Progress made during the period 1 July 2009 – 30 June 2010 is described below. Specifically, the majority of ***Task 10*** is now completed; preparation of manuscripts (***Task 11)*** is in progress.

The project is currently operating under an approved no-cost extension to 30 June 2011 to complete the remaining project tasks.

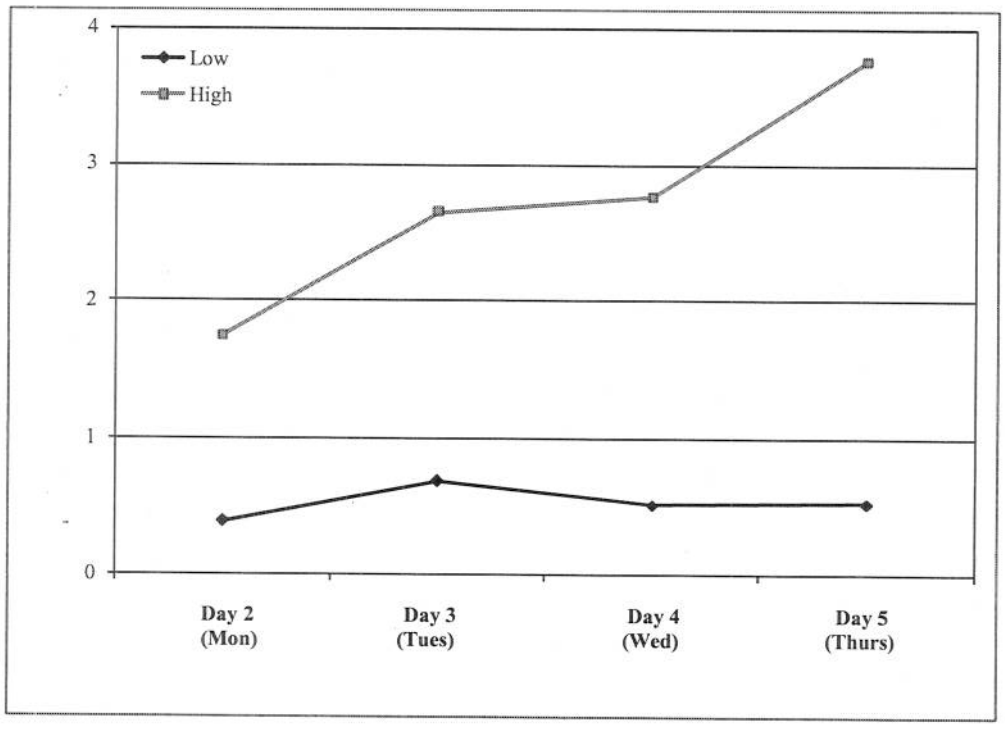
As summarized in previous years' Annual Reports, Tier I phase data collection conducted in Jan 2007 involved a total of 24 participants at one Air Force base (AFB). Tier II phase data collection was carried out with a total of 74 participants between January-April 2008 at three different US Air Force bases (AFB1: 25 Jan- 1 Feb 2008; AFB2: 28 Mar-4 Apr 2008; AFB3: 18-25 April 2008). In brief, the study design involved recruiting participants from higher and lower exposure group categories (based on *a priori* job-type activities). Each participant was asked to participate in the study over a period of 6 work days, with his/her study participation starting on a Friday afternoon (Day 1) and continuing Monday morning through Friday morning (Days 2-6) the following week. Biological and/or environmental samples of JP8 exposure were collected from each participant every day, along with daily questionnaires and scheduled neurobehavioral task and postural sway testing. Laboratory analyses of all collected samples were completed by Dec 2009.

***Task 10:* Complete data analyses of exposure-outcome hypothesis relationships (Tier II)- In PROGRESS (with most all completed).**

As described in last year's Annual Report, the data management steps to integrate the Tier II in-person questionnaire, neurobehavioral performance and postural sway data and environmental and biological sample results as needed for data analyses procedures, have been completed.

Summary of analyses focused on exposure. Over the past year, linear mixed-effects modeling analyses have been performed to examine relationships between JP8 inhalation/air exposure, job tasks, and urinary metabolites over the workweek. One additional aspect of analyses has been to depict the considerable variation in exposure levels over a workweek (**Figure 1**).

Figure 1. Levels of personal (geometric mean) air THC concentration (mg/m3) over the work week, by *a priori* exposure (high, low) groups



Summary of findings:

- *Air levels of JP8 components were significantly higher among the hypothesized higher exposure group*
- *Total hydrocarbon(THC) air levels were significantly different among job task categories*
- *THC levels were significantly correlated with naphthalene levels in air*
- *Post-shift urinary 1-naphthol levels were significantly higher than pre-shift levels among the high exposure group*
- *THC air levels were significantly associated with urinary 1-, and 2-naphthol levels*
- *Naphthalene air levels were slightly stronger predictors of urinary 1-,2-naphthol levels than THC air levels*

Summary of analyses focused on exposure-outcome relationships. Primary data analyses set to examine the hypothesized exposure-outcome relationship between measured exposure levels and neurological functioning (i.e., neurocognition, balance) have been completed, with additional data analyses for final manuscript preparations in progress.

In summary, to examine the study hypotheses regarding occupational exposure to JP8 and neuropsychological functioning as part of Tier II, neuropsychological testing was conducted at the end of shift on the first day of the study (Day 1 Battery) and subsequently on at the start of shift on Day 2, Day 4, and Day 6 (Repeated Day Battery). The neurobehavioral task batteries **(Table 2A & B)** were designed to be feasible in a field study environment, given time and environmental constraints, and to provide appropriate and reliable measurements of performance in a repeat testing scenario.

Table 2A. Neuropsychological Day 1 Battery: Task Descriptions

Test	Domain Assessed	Outcomes Measured	Possible Score Range	Reference
Shipley Vocabulary	General academic ability	# of correct responses	0-40	Shipley, 1946
Hooper Visual Organization Test	Visuospatial ability	# of correct responses	0-30	Hooper , 2004
Hopkins Verbal Learning Test: Total Recall	Verbal learning	# correct, sum of trials 1-3	0-36	Brandt, 1991
Delayed Recall	Verbal memory	Number correct, trial 4	0-12	
Retention (%)	Verbal memory	Delayed Recall/(Higher of recall score 2 or 3)*100	0-100	
Recognition Discrimination Index	Verbal memory	Total True Positives – Total False Positives	0-12	

To increase experimenter reliability and facilitate administration and data management efficiency, several tasks are administered in a computer-assisted format (tasks from the Automated Neuropsychological Assessment Metrics (version 4, ANAM4) test battery, C-SHOP-ANAM4 2007). Other traditional paper-pencil neuropsychological tasks that focus on particular functional domains of importance, but not tapped via the computer-assisted tasks, were

included. Also, at the time of each neuropsychological test session, participants were administered the Positive and Negative Affect Scale (PANAS) to assess current mood state, and completed the ANAM4 Sleepiness Scale. On Day 1, all participants were administered trial 1 of the Test of Memory Malingering (TOMM), which is a simple 50-item visual memory test assessing cognitive engagement. It was administered for the purpose of excluding persons from the analyses who exhibit low levels of engagement in the objective cognitive tests. Previous research examining the sensitivity and specificity of the TOMM indicates that a score below 38 on trial 1 of the TOMM suggests insufficient task engagement; in this study, no participant scored below 38.

Table 2B. Repeated Day Battery*:Task Descriptions

Test	Domain Assessed	Outcomes for Analyses	Possible Score Range	Reference
ANAM4 Match to Sample	Visuospatial ability, visual memory	Throughput	-	Vincent et al., 2008 others
ANAM4 Simple Reaction Time	Attention, psychomotor ability	Throughput	-	
ANAM4 Standard CPT	Sustained attention	Response time # NR (Omission) errors # FP (Commission) errors	-	
ANAM4 Finger Tapping Dominant hand Non-dominant hand	Psychomotor speed	Mean # of taps, from 2 trials	-	
Auditory Consonant Trigrams – 9 s delay – 18 s delay – 36 s delay	Executive function, memory	 # correct # correct # correct Total correct	 0-15 0-15 0-15 0-45	Stuss et al, 1987; Strauss et al, 2006
WAIS3 Digit Span Forward Backward	Attention	# correct spans	0-16 0-14	Wechsler, 1981; Strauss et al, 2006
Grooved Pegboard Dominant hand Non-dominant hand	Fine motor abilities	Time to complete	0-300 0-300	Matthews and Klove, 1964; Strauss et al, 2006

***administered on Study Days 2, 4, 6 (Mon, Wed, Fri mornings)**

Summary of findings:

- *Only subtle differences in neurocognitive functioning are noted in those persons with greater than 10 years of Air Force service and those currently working in jobs with higher JP8 jobs (analyses of performances on the Day 1 battery)*
- *Overall, performance on most all cognitive task performances was observed to improve over the workweek*

- *Significant patterns of association between JP8 exposure and cognitive performance over the workweek were observed on tasks involving sustained attention (analyses of performances on the Repeated Day battery), but not observed on other tasks*
- *Minimal to no significant changes in balance parameters are observed over a work shift*

Task 11: **Prepare Final Report and manuscript(s)-In PROGRESS.**
Throughout this past year, the study team has been working on preparing manuscripts for publication, following our overall paper topic (based on study hypotheses) plan and sequence for submission.

Specifically, as the Tier II phase builds from information garnered in Tier I phase, we have focused on completing submitting for publication consideration a set of 3 papers from Tier I and an overview report (that describes the OJENES project research approach and the knowledge gaps it addressing) (**Table 3**). In addition, 5 manuscripts from Tier II are in progress.

Table 3. List of OJENES manuscripts and current status.

	Title or [Topic]	Manuscript Status
Overview	The Occupational JP8 Exposure Neuroepidemiology Study (OJENES): Repeated workday exposure and central nervous system functioning among Air Force personnel.	Under internal review
Tier I		
	Inhalation exposure to jet fuel (JP8) among US Air Force personnel	In press, JOEH
	Urinary biomarkers of occupational jet fuel (JP8) exposure among Air Force personnel	Submitted
	Biomarkers of exposure to jet fuel (JP8) in exhaled breath among Air Force personnel	Under final internal review
Tier II		
	Characteristics of jet fuel inhalation exposure among US Air Force personnel	Under internal review
	[JP8 exposure histories and neurocognitive functioning]	In preparation
	[Repeated workday exposure to JP8 and changes in neurocognitive performances over a workweek[	In preparation
	[Workday exposure to JP8 and balance]	In preparation
	[Repeated measures of urinary biomarkers of occupational jet fuel (JP8) exposure]	In preparation

°Additional analyses planned include examination of glutathione-S-transferase (GST) enzyme polymorphisms and association with JP8 exposure and central nervous system functioning

Also, in the past year, two abstracts from ISEE (Aug 2009, Dublin Ireland) meeting presentations have been published (and are included in the Appendix):

- Smith KW, Proctor SP, McClean MD. Relationships between inhalation exposure, urinary and end exhaled-breath biomarkers among jet fuel exposed Air Force personnel. Epidemiology 2009; 20:S167. (from Tier I results)
- Rodrigues EG, Merchant-Borna K, Smith KW, Proctor SP, McClean M. Characterization of jet fuel inhalation exposure and urinary metabolites in US Air Force personnel. Epidemiology 2009; 20:S60. (from Tier II results)

Key Research Accomplishments

Below is a bulleted list of the accomplishments over this study period:

- The Exposure Assessment Methodology Working Group, Neurology Working Group, and the Data Management and Logistics Working Group met on regular (monthly to quarterly) bases to discuss on-going data analyses and manuscript preparations.
- Final analyses of biological samples collected during Tier II and sent to CDC have been completed.
- The abstract focused on the Tier I air, urine, and exhaled breath findings was presented at the International Society of Environmental Epidemiology (ISEE) meeting (August 2009) in Dublin, Ireland.
- The abstract focused on the Tier II air and urine findings was presented at the ISEE meeting (August 2009) in Dublin, Ireland.
- A manuscript describing Tier I has been accepted for publication in the Journal of Occupational and Environmental Hygiene (JOEH): Smith KW, Proctor SP, Ozonoff A, McClean MD. "Inhalation Exposure to Jet Fuel (JP8) Among U.S. Air Force Personnel.
- Several other Tier I phase manuscripts and Tier II manuscripts are in various stages of preparation and submission (see above listing in **Table 3**).
- Continuing Review Reports have been reviewed and approved by the USARIEM Human Use Research Committee (Feb 2010), AFRL/Wright Site IRB (March 2010) and VA Boston Healthcare System (Jan 2010).
- The PI and several members of the study team have visited and/or communicated with AFBs involved (Fall 2009) to provide briefings of the preliminary results from the Tier II phase.

Reportable Outcomes

1. Reports, manuscripts, abstracts

- See **Table 3** above for status of all project-specific papers and abstracts.

- Additionally, colleagues at CDC presented on the volatile organic compound (VOC) analysis method used in this study at the 2010 Society of Toxicology meeting in Salt Lake City, March 2010.
    - Alwis KU, Blount BC, Sheppard A, Proctor SP, Ashley DL. Simultaneous analysis of eleven VOC metabolites in human urine. Abstract published in The Toxicologist 2010; 114: 277-278.

2. Degrees and research training opportunities

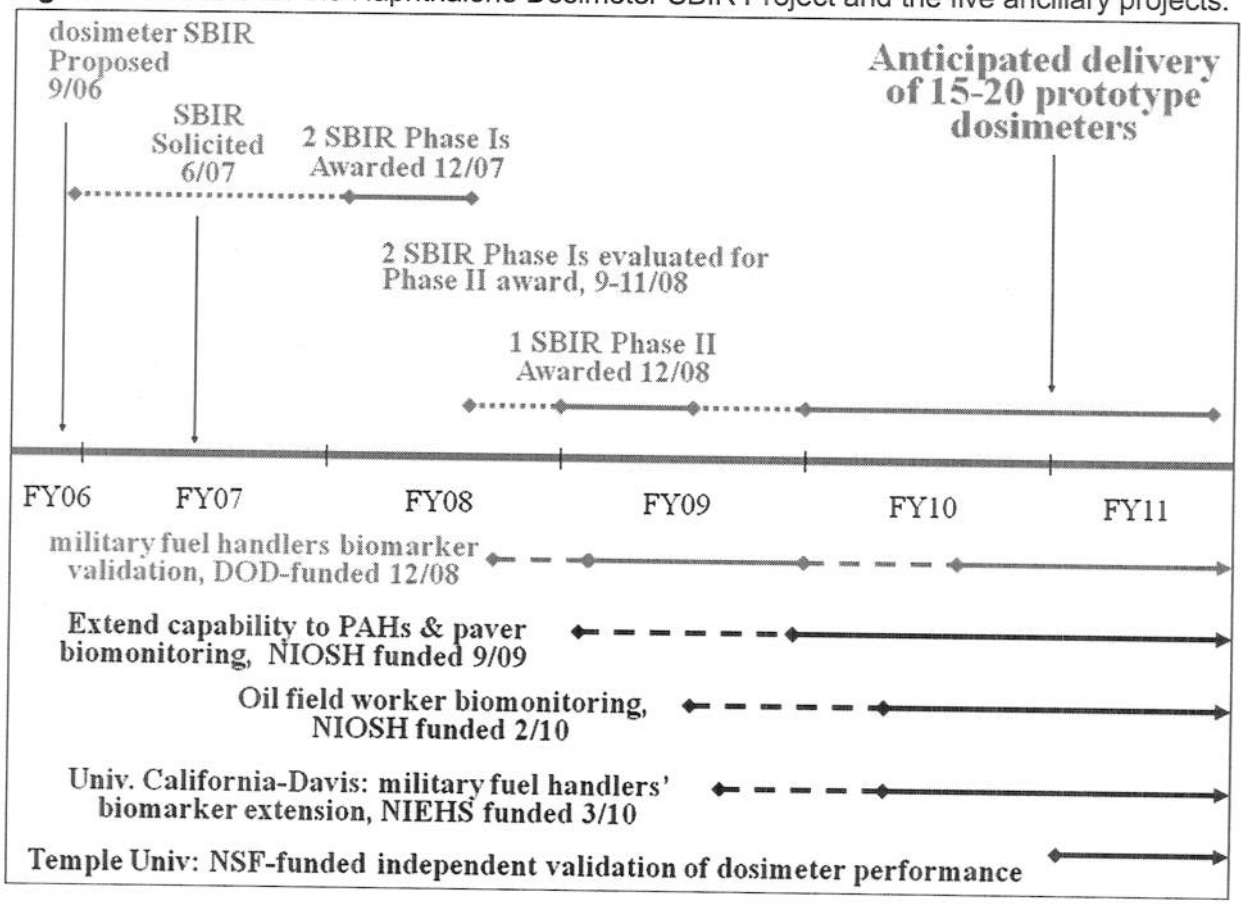
- Several Boston University School of Public Health (BUSPH) students (and a recent doctoral degree graduate, May 2010) have served as integral members of the Tier II field study team and continue to conduct data analyses and prepare manuscripts for journal submissions.

3. Collaborative funding applications related to work supported by this award

- The PI is involved (at co-PI level) in a new project 'Measuring Naphthalene and Biological Markers of Exposure among Military Fuel-Worker Personnel: The Naphthalene Dosimeter Field Validation Study'. The goals of this study are to test and validate the ability of the Army Research Office- Small Business Innovative Research (SBIR) Program- developed naphthalene (wearable) dosimeter prototype instruments to efficiently and accurately measure workplace levels of naphthalene in (near) real time and to examine the extent to which the naphthalene exposure levels measured with the newly developed instrument correlate with environmental and biological indices of exposure to naphthalene. The project managers on the naphthalene dosimeter SBIR project are Dr. Janis Hulla, US Army Corps of Engineers and Dr. Micheline Strand, Army Research Office. This research validation study is funded by the Office of the Secretary of Defense (OSD) Chemical and Materials Risk Management Directorate to the Army Research Office (Dr. Strand, Contract Officer's Representative) and is implementing this project through the CDC Foundation, the Army Corps of Engineers (Dr. Hulla, Principal Investigator), NIOSH (Dr. Snawder, Lead Investigator) and USARIEM (Dr. Proctor, Lead Investigator).

- In addition to the Army-funded SBIR project, there are currently five ancillary projects funded by the DOD, National Science Foundation, NIOSH (two) and National Institute of Environmental Health Sciences (**Figure 2).** The human subject validation research project is depicted in blue.

Figure 2. Timeline for the Naphthalene Dosimeter SBIR Project and the five ancillary projects.



- An overview of DoD Naphthalene Dosimeter research has been presented in several Defense Knowledge Online website locations. A poster summarizing this work was presented at the 2010 Society of Toxicology meeting in Salt Lake City, March 2010.
    - Hulla, JE, Snawder JE, Proctor SP, Chapman GD. DoD impact assessment and management of naphthalene-related risks. Abstract published in The Toxicologist 2010; 114: 400.

4. Related projects and collaborations initiated

- The PI is currently serving as a member of the Naphthalene Dosimeter Advisory Group, chartered from the OSD, *Chemical and Materials Risk Management Directorate*

- The PI and Boston University Exposure Assessment team have provided collaborative assistance to the NIOSH Biomonitoring Research Team (Dr. John Snawder) in preparation of a new initiative to study and characterize workplace exposures via direct read monitors.

- The PI was invited to attend the MOMRP Pulmonary Health Task Working Group Meeting in June 2010 in Frederick, MD. This meeting and other DoD initiatives have focused increased attention on exposure assessment and biomarker efforts under operational conditions.

- The PI was invited to attend the scientific symposium entitled "Assessing Potentially Hazardous Environmental Exposures among Military Populations" at USUHS in May 2010. The meeting was sponsored by the Armed Forces Health Surveillance Center (AFHSC) and the Uniformed Services University of the Health Sciences (USUHS).

Conclusions

There has been substantial progress over this funding period. With the completion of the final stage of the project (publication of study findings), the study will provide important occupational health and exposure assessment information concerning JP8 in repeated workday settings.

As stated in the recent report (National Research Council, 2003), field research studies that combine the in-depth assessment of on-the-job exposure levels with concurrent assessment of adverse health effects are needed and will contribute significantly to the knowledge of the subclinical effects of both acute and chronic exposure to occupational solvent exposures.

References

Subcommittee on Jet-Propulsion Fuel 8, Committee on Toxicology, National Research Council. (2003). *Toxicologic Assessment of Jet-Propulsion Fuel 8*. Washington, D.C.: The National Academies Press.

Appendix

ISEE 2009 Abstracts

- Smith KW, Proctor SP, McClean MD. Relationships between inhalation exposure, urinary and end exhaled-breath biomarkers among jet fuel exposed Air Force personnel. Epidemiology 2009; 20:S167.
- Rodrigues EG, Merchant-Borna K, Smith KW, Proctor SP, McClean M. Characterization of jet fuel inhalation exposure and urinary metabolites in US Air Force personnel. Epidemiology 2009; 20:S60.

SOT 2010 Abstracts

- Alwis KU, Blount BC, Sheppard A, Proctor SP, Ashley DL. Simultaneous analysis of eleven VOC metabolites in human urine. The Toxicologist 2010; 114: 277-278.
- Hulla, JE, Snawder JE, Proctor SP, Chapman GD. DoD impact assessment and management of naphthalene-related risks. The Toxicologist 2010; 114: 400.

ISEE-0485

Association Between Cotinine and Metals in Maternal and Cord Blood in Non-Smoking Mothers

Ting-Wen Wen, and Pau-Chung Chen, *Institute of Occupational Medicine and Industrial Hygiene, Collage of Public Health, National Taiwan University, Taipei, Taiwan.*

Background and Objective: Environmental tobacco smoke (ETS) contains over 4000 compounds, including numerous heavy and trace metals such as arsenic, lead, cadmium, and selenium. Smokers have been reported to have higher blood lead and cadmium levels than do nonsmokers. The objective of this study was to explore the association between cotinine and metals in maternal and umbilical cord blood in non-smoking mothers.

Methods: The study population consisted of 328 postpartum women collected from four hospitals and clinics in northern Taiwan. We interviewed them by a structured questionnaire after delivery and collected maternal and umbilical cord blood at birth. Cotinine in blood as an indicator of ETS was analyzed by using HPLC-MS/MS and the metals were analyzed by Agilent 7500 C inductively coupled plasma mass spectrometry (ICP-MS). We examined the association between cotinine and $\log_{10}$ transformed metal levels by linear regression models.

Result: After adjusting for maternal age and education, there were negative association between cadmium ($\beta \pm SE = -0.00005 \pm 0.0011$, P-value = 0.961), antimony ($\beta \pm SE = -0.00026 \pm 0.0007$, P-value = 0.689) and barium ($\beta \pm SE = -0.00414 \pm 0.0025$, P-value = 0.095) and cotinine in maternal blood. In umbilical cord blood, a negative association was found for antimony ($\beta \pm SE = -0.00113 \pm 0.0005$, P-value = 0.020) while positive associations were shown for thorium ($\beta \pm SE = 0.00237 \pm 0.0011$, P-value = 0.028) and uranium ($\beta \pm SE = 0.00307 \pm 0.0015$, P-value = 0.046).

Conclusions: Although cotinine were associated with some metals in blood, environmental tobacco smoke may not be the major source of metals in the non-smoking population.

ISEE-0488

Relationships Between Inhalation Exposure, Urinary and End Exhaled-Breath Biomarkers Among Jet Fuel Exposed Air Force Personnel

Kristen Smith,*† Susan Proctor,† and Michael McClean,* **Boston University School of Public Health, Boston, MA, United States; and †US Army Research Institute of Environmental Medicine, Natick, MA, United States.*

Background and Objective: Jet propulsion fuel 8 (JP8) and similar jet fuels are widely used by the US military and commercial airline industry, resulting in widespread occupational exposures that could potentially cause adverse neurological health effects. The objectives of this study were to characterize JP8 exposure by examining exhaled-breath biomarkers between a priori designated exposure groups and assessing relationships with both inhalation exposure and urinary biomarkers.

Methods: Air Force (AF) personnel (n = 24) were recruited from an active USAF base into low, moderate, and high a priori designated exposure groups. Exhaled-breath samples were collected over three consecutive work-days and analyzed for benzene, toluene, ethylbenzene, xylene (BTEX), hexane, and naphthalene. Urine samples were collected concurrently and analyzed for 1- and 2-naphthol. Breathing-zone air samples were collected over the work-shift and analyzed for total hydrocarbons (THC), BTEX, and naphthalene. Linear mixed effects models were used to evaluate the exposure data.

Results: The geometric mean post-shift exhaled-breath concentrations for participants in the low, moderate, and high exposure groups were <6.5 ug/m^3, 9.0 ug/m^3, and 10.4 ug/m^3 for hexane; results for BTEX were similarly ordered. Exhaled-breath naphthalene concentrations were excluded from the analyses due to a low limit of detection. In post-shift exhaled breath samples, exposure group was a significant predictor of hexane ($P = 0.01$), ethylbenzene ($P < 0.0001$), m-/p-xylene ($P < 0.0001$), and o-xylene ($P < 0.0001$) with levels increasing across the low to high exposure groups. In pre-shift exhaled breath samples, exposure group was also a significant predictor of ethylbenzene ($P = 0.01$), m-/p-xylene ($P = 0.005$), and o-xylene ($P = 0.01$). Post-shift exhaled-breath hexane and BTEX measurements were weakly to moderately correlated with THC measured in personal air (r = 0.1–0.5) and moderately correlated with post-shift urinary 1- and 2-naphthol (r = 0.4–0.6).

Conclusion: Exhaled-breath concentrations increased across the low to high a priori designated exposure groups and were correlated with urinary biomarkers.

ISEE-0489

Effects of Household Use of Cleaning Products on Birth Weight

Lidia Casas,*† Jan Paul Zock,*† Mar Álvarez-Pedrerol,*† Mònica Guxens,*† and Jordi Sunyer,* **Centre de recerca en epidemiologia ambiental (CREAL), Barcelona, Spain; and †Institut Municipal d'Investigació Mèdica (IMIM), Barcelona, Spain.*

Background and Objective: Associations between frequent use of household chemicals during pregnancy and wheezing and diarrhoea in offspring have been reported, but there are no studies on the effects of such exposures on birth weight (BW). The aim of this study is to assess the association between the use of domestic cleaning products during pregnancy and BW.

Methods: In the Spanish longitudinal INMA-Sabadell birth cohort study, 619 pregnant women were followed from the first trimester of pregnancy until delivery. Birth outcomes were obtained from clinical records of 617 newborns (≥34 weeks of gestation). The use of cleaning products in the home was obtained from an interviewer-led questionnaire administered at the third trimester of pregnancy. Associations between the use of cleaning products and BW were evaluated using multivariable linear regression models adjusting for sex, gestational age, mother's height, weight and number of previous pregnancies.

Results: The median BW was 3288g (interquartile range 2970 to 3520g). The most commonly used cleaning products were glass cleaners (77%), bleach (74%), furniture polishes (42%) and ammonia (25%). Women who used bleach had newborns with a higher BW (mean difference 87g; 95%CI 17 to 152). Similar results were found for ammonia (mean difference 60g; 95% CI −11 to 130). The association between bleach and BW remained apparent after additional adjustment for tobacco smoking during pregnancy, maternal education and employment in cleaning work (mean difference 71g; 95%CI: −3 to 146) and showed a dose-related trend (mean difference 62 and 96g for frequency of cleaning ≤1 and >1 time/week, respectively). Other products were not associated with BW.

Conclusions: Household use of bleach during pregnancy was associated with a higher BW. We hypothesise that a higher degree of disinfection of the living environment could be beneficial for foetal development.

ISEE-0492

Initial Enrollment of Asthmatic Children in a Woodstove Intervention Study

Curtis Noonan, Tony Ward, and Emily Weiler, *University of Montana, Missoula, MT, United States.*

Background: This study utilizes in-home woodstove interventions to assess the impact of indoor biomass smoke on asthmatic children. Initial enrollment efforts, methodologies, and the descriptive characteristics of the first cohort of participants are described here.

Methods: Asthma screening surveys were administered to school children (n = 1,185) to identify subjects. Baseline indoor air sampling and health measures were conducted during the winter of 2008/09. Air sampling

ISEE-0487

Quantification of Short Term Effects of Pollen Counts and Sentinel Botanic Garden Observations on Pollinosis Symptoms: A French Panel Study

Gaëlle Pedrono,* Christelle LE Grand,* Marie-Thérèse Guillam,* Alain Meunier,† Daniel Riviere,† Claude Figureau,‡ Michel Thibaudon,§ Angéline Vinat,* Yann Dubreil,¶ Dominique Chevallier,¶ Laurent-Charles Antoine,¶ Odile Morin,¶ and Claire Segala,* **SEPIA-Santé, BAUD, France; †DRASS Pays de Loire, NANTES, France; ‡Ville de Nantes, NANTES, France; §Réseau National de Surveillance Aérobiologique, LYON, France; and ¶AEROCAP44, NANTES, France.*

Background and Objective: In recent decades, numerous epidemiological studies investigating the change of prevalence of pollinosis showed an increase in the occurrence of this disease all over the world. In order to explain this phenomenon and to build a specific prevention tool, a sentinel botanic garden was realized in Nantes city (France). The objective of this study was to explore short term relationships between pollinosis symptoms and either grass pollen counts or pollen observations in this garden.

Methods: 81 volunteers suffering from pollinosis and living in Nantes metropolis area were recruited by allergy physicians. The panel took place while the grass pollen season (March to July 2007). Daily number of plants in flower was collected by botanists and daily pollen counts were measured by a central stationary sampler. Daily symptoms of rhinoconjunctivitis (eye, nose, throat, respiration) were reported in a diary by volunteers as well as pollinosis treatment intake.

Marginal (GEE) and mixed models were realized to explore short term effects of pollen covariates on pollinosis symptoms and treatment intake. Models were adjusted for confounding variables (time trend, meteorology, pollution levels) and took lags and autocorrelation into account.

Results: The response rate was excellent: 97%. Most of the volunteers were treated by antihistaminic and immunotherapy. A positive and significant association between prevalent pollinosis symptoms and grass pollen counts was shown for nose (OR = 1.043; 95%CI [1.026–1.060]) and eyes (OR = 1.035; 95%CI [1.018–1.052]) symptoms. Short term effects of pollen observations were also observed in the period before the pollen grass peak for total symptoms (OR = 1.02; 95%CI [1.00–1.05]).

Conclusion: Short term effects of grass pollen on pollinosis symptoms were clearly showed and quantified. Those results provide information for better prevention and care of pollinosis and have contributed to the establishment of a new panel study realized in 2009.

ISEE-0491

Characterization of Jet Fuel Inhalation Exposure and Urinary Metabolites in U.S. Air Force Personnel

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Background and Objective: Jet propulsion fuel-8 (JP-8) is the primary jet fuel used by the U.S. military, collectively consuming about 2.53 billion gallons annually. Previous reports suggest that JP-8 is potentially toxic to the immune, respiratory, and nervous systems. The objectives of this study were to evaluate inhalation exposure to JP-8 as well as absorption of JP-8 constituents among U.S. Air Force (USAF) personnel while performing job-related tasks.

Methods: Seventy-three full-time USAF personnel from three active bases were categorized a priori as having low (n = 35) or high (n = 38) exposure to JP-8 based on job title and tasks. Personal air samples were collected from each participant during four consecutive workdays using air pumps and sorbent tubes.

Using gas chromatography/mass spectrometry, charcoal sorbent tubes were analyzed for benzene, ethylbenzene, toluene, xylenes, and total hydrocarbons (THC) while Chromosorb® tubes were analyzed for naphthalene. Pre- and post-shift urine samples were also collected from each worker each day and analyzed for 1- and 2- naphthols, 2-, 3-, and 9-hydroxyflourene, 1-, 2-, 3-, and 4-hydroxyphenanthrene, and 1-hydroxypyrene. Linear mixed-effects models were used to explore the association between inhalation exposure and post-shift urinary metabolites, adjusting for creatinine and pre-shift urinary concentrations.

Results: THC air concentrations were significantly different between the exposure groups (2.6 vs. 0.5 mg/m^3, $P < 0.0001$). Similar differences were observed for the other analytes measured in air. Among the high exposure group, post-shift urinary 1- and 2-naphthol levels were significantly higher than pre-shift levels (both $P < 0.05$). Inhalation exposure to THC was significantly associated with post-shift urinary 1-naphthol ($\beta = 0.21$, $P < 0.0001$), 2-naphthol ($\beta = 0.11$, $P = 0.0006$) and 2-hydroxyflourene levels ($\beta = 0.08$, $P = 0.005$). Naphthalene air concentrations displayed similar significant associations with post-shift urinary 1-naphthol ($\beta = 0.26$, $P < 0.0001$) and 2-naphthol levels ($\beta = 0.13$, $P < 0.0001$).

Conclusion: USAF personnel experience inhalation exposure to JP-8 which is associated with absorption of JP-8 constituents while performing normal job-related tasks.

ISEE-0498

A Panel Study on Epigenetics, Markers of Oxidative Stress, and Lung Function Among Children with Respiratory Disease Exposed to Industrial Air Pollution

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Objectives: To study DNA methylation, exhaled nitric oxide (FeNO), lung function (FEV1) in children with respiratory symptoms exposed to industrial air pollution.

Methods: A panel study of 39 children aged 8–11 years followed on 2007/12–2008/4 was conducted in Valle-del-Mela (Sicily-Italy), a High Risk Area (55504 inhabitants) with oil refineries and energy plants. Symptomatic children were screened by modified ISAAC questionnaire (2506, 89.5% responders). The 39 selected children were divided into 9 groups matched by school, monitored for 7 consecutive days. DNA Methylation was measured on nasal mucosa cells collected by swab, twice per subject on day fourth and seventh of the same week. Personal $PM_{2.5}$ active, NO_2, SO_2 passive sampling were done on one child witness of a 4-child group. Ambient $PM_{2.5}$ monitor, meteo station, passive NO_2 SO_2 samplers in 21 schoolyards were used. Diaries filled in by parents recorded symptoms, therapy, indoor sources. Data were analyzed with mixed models controlling for confounders.

Results: Average daily ambient $PM_{2.5}$ was 23.0 μg/m^3, weekly ambient SO_2 over 20 μg/m^3 in three locations. Average daily (90 percentile) personal $PM_{2.5}$ was 44.5 (86.6), SO_2 17.7 (32.8). Effect measures were expressed for 10 μg/m^3 increase of pollutant concentration.

We found FEV_1 reduction −4.3% (90% Confidence Interval −6.1; −2.6%) for SO_2 lag2 ($P < 0.01$), FeNO increment 10.8% (3.2–18.4%) for SO_2 lag01 ($P = 0.022$); a decrease −1.0% of global DNA Methylation (Alu elements 90% CI −2.0; −0.6%) and −4.1% of iNOS (−7.8; −0.4%) for SO_2 lag12; −1.8% (−3.0; −0.6%) of iNOS for $PM_{2.5}$ lag12. DNA Methylation of interleukin 6 position was reduced when FEV_1 was

PS 1297 INVESTIGATION OF THE NEUROTOXIC MECHANISMS INVOLVED IN BETA-AMYLOID DEPOSITION IN PSAPP MICE.

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Introduction: Amyloid-beta is endogenously formed neuronal peptide which has been proved to have a causal relationship with neurodegeneration in Alzheimer's disease (AD). MAP kinase, CREB, ERK and pCREB have played a vital role in memory regulation. The present study employed PSAPP mice expressing the "Swedish" amyloid precursor protein and M146L presenilin-1 (PSAPP) mutations to study the cellular mechanisms and biomarkers involved in Aβ toxicity in relation to the loss of memory. Experimental Procedures: PSAPP mice and non-transgenic controls (eight months old) were subjected to behavioral and biochemical studies. Brains were dissected, hippocampus and cortex were removed. Behavioral experiments such as Y-maze and open field were performed along with Aβ deposition (1-40 and 1-42). Enzymatic activity of beta secretase as well as the alteration in the cellular signaling pathways (ERK MAP kinase, STAT and CREB pathways were analyzed by multiplex microbeads method. ANOVA and Dunnett's test were used to compare the results with non-transgenic mice. Results and Conclusion: This study reveals the alterations in behavioral and cellular processes that occur due to Aβ deposition in PSAPP transgenic mice. PSAPP mice exhibited significant Aβ deposition (1-40 and 1-42) and behavioral deficits (y-maze and Open field) compared to the control non-transgenic mice. Beta-secretase activity was significantly increased in the PSAPP mice in the cortex and hippocampus. The kinases such as ERK MAP kinase, JNK, p70S6 kinase exhibited down regulation in the transgenic animals. Other cellular biomarkers such as STAT5, STAT and CREB also showed the same trend. Administration of exogenous Aβ peptide has also shown to induce characteristic neurodegeneration in the hippocampus. However, the cellular mechanisms differ as compared to the endogenous deposition of Aβ. The study of these cellular processes and their changes can divulge important targets that could be utilized for newer drug discovery. Acknowledgement: Alzheimer's Association grant (NIRG-08-91816)

PS 1298 NEONATAL EXPOSURE OF MALE RATS TO BISPHENOL A IMPAIRS EXPRESSION OF SERTOLI CELL JUNCTIONAL PROTEINS IN THE TESTIS.

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Sertoli cell junctional proteins (SCJP) (viz. adhesion, gap and tight junctions) are important for spermatogenesis and perturbations in expression of these proteins are associated with impairments in process of sperm production. Bisphenol A (BPA) is an endocrine disrupter that has been associated with impaired spermatogenesis. However the mechanistic basis of impaired spermatogenesis is unknown, whether BPA is a Sertoli cell toxicant has not yet been fully investigated. The present study was undertaken to decipher the effects of neonatal exposure of male rats to BPA on the testicular expression of SCJP during development. Neonatal male rats were s.c injected with 2.4 µg/day (300 µg/kg bw) of BPA in sesame oil from postnatal day 1-5 and controls received vehicle. Immunohistochemical localization for Connexin 43 (Cx-43, gap junctional), Zona Occludin-1 (ZO-1, tight junctions) and N-cadherin (adherens junction) was carried out on testicular tissue sections obtained from PNDs 15, 30, 45 and 90 of rats exposed to the lowest dose of BPA(2.4 µg/day) that impaired fertility. A significant reduction in the expression of Cx-43 (PND 45 and 90) and increases in the expression of N-cadherin (PND 45 and 90) and ZO-1(PND 90) were observed in the testes of rats exposed neonatally to BPA. Interestingly, there was an altered expression pattern of Cx43 amongst the sloughed cells in the testes of the experimental rats as compared to controls. Neonatal exposure of BPA to rats has the potential to induce perturbations in SCJP. These perturbations may be one of the contributing factors that lead to impairments in spermatogenesis in the exposed animals and can be used as potential biomarkers to study BPA- induced effects on testes.

PS 1299 DETECTING BIOMARKERS OF CHRONIC ARSENIC EXPOSURE BY USING SELDI-TOF-MS PROTEIN CHIP TECHNOLOGY.

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Background: Chronic exposure to high levels of inorganic arsenic that is naturally present in drinking water in certain geographic regions has become a major public health concern in China. In this study we used surface-enhanced laser desorption/ionization time-of-flight mass spectrometry (SELDI-TOF-MS) to determine if arsenic exposure in drinking water could induce serum protein changes and to identify new protein biomarkers for chronic arsenic exposure. Methods: A total of 178 subjects were selected in three groups based on arsenic exposure levels in drinking water (3.2±2.5 µg/L, 22.1±4.7 µg/L, and 177.6±23.8 µg/L, respectively). Serum proteomic profiles were analyzed by SELDI-TOF-MS with a CM10 Protein Chip. Diagnostic model was constructed by decision tree algorithm in a training set with 120 subjects and validated in a testing set with other 58 subjects. Results: Relative intensities of 41 protein peaks were found differently among three groups. A panel of five proteins with mass-to-charge ratio (m/z) of 2872.48, 6121.42, 7580.58, 9432.56 and 5552.66 was selected to build the diagnostic model. Among these markers, the 2872.48 Da and the 7580.58 Da were significantly up-regulated or down-regulated only in the group of subjects exposed to 177.6±23.8 µg/L of arsenic. The 6121.42 Da and the 5552.66 Da were significantly down-regulated in the groups with arsenic exposure levels of 22.1±4.7 µg/L and 177.6±23.8 µg/L. The 9432.56 Da content was the lowest in the group exposed to 22.1±4.7 µg/L of arsenic. The power to detect differences among three groups in the testing set was evaluated with the sensitivity of 80.00%-86.67%, and the specificity of 86.67%-95.35%. Conclusion: Exposure to 22.1±4.7 µg/L of arsenic in drinking water is enough to cause changes in serum protein profiles. This proteomic technology showed very promising in detecting levels of arsenic exposure and discovering new biomarkers.

PS 1300 EFFECTS OF ORAL ADMINISTRATION OF PIOGLITAZONE, SODIUM SACCHARIN OR SODIUM O-PHENYLPHENATE ON THE EXPRESSION OF ONCOMODULIN IN THE BLADDER EPITHELIUM OF MALE F344 RATS.

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Currently there are no reliable markers for early detection of urinary bladder cancer. Recent studies have shown that expression of the oncomodulin gene is increased in urothelium from rats treated with various bladder carcinogens. Pioglitazone is a PPARγ agonist which induces rat bladder tumors. We administered pioglitazone in 0.5% methylcellulose (MC) intragastrically (i.g.) to evaluate the level of oncomodulin expression in F344 rat urinary bladder epithelium. Sixty male F344 rats were randomized into 4 groups of 15 rats each and treated for 4 weeks with: 1) control diet and daily MC i.g.; 2) control diet and 16 mg/kg pioglitazone in MC i.g.; 3) diet containing 7.5% sodium saccharin (NaSac) and MC i.g.; or 4) diet containing 2.0% sodium o-phenylphenate (NaOPP) and MC i.g. RT-PCR was employed to detect expression of oncomodulin in the urothelium. Light microscopy, SEM, and immunohistochemical detection of BrdU were used to examine cytotoxic and proliferative urothelial effects. Expression of oncomodulin was significantly increased in NaSac or NaOPP-treated groups compared to controls, but not in the pioglitazone group. All test chemicals induced superficial necrosis by SEM and increased BrdU labeling index indicative of increased cell proliferation. In vitro, PPARγ agonists induced differentiation in rat urothelial cells (MYP3), decreased proliferation, and decreased oncomodulin expression. Unlike NaSac and NaOPP, pioglitazone did not induce an increase in oncomodulin expression, possibly related to its competing effects of, 1) indirectly increasing urothelial proliferation by inducing production of urinary solids, and 2) decreasing proliferation due to direct effects on urothelial PPARγ.

PS 1301 SIMULTANEOUS ANALYSIS OF ELEVEN VOC METABOLITES IN HUMAN URINE.

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Volatile organic compounds (VOCs) are ubiquitous in the environment, originating from many different natural and anthropogenic resources, including tobacco smoke. Long-term exposure to certain VOCs may increase the risk for cancer, birth defects, and neurocognitive impairment. Therefore, VOC exposure is an area of significant public health concern. We developed a reversed-phase high performance liquid chromatography coupled with electro-spray ionization tandem mass spectrometry (LC-ESI/MSMS) method to quantify urinary VOC metabolites as biomarkers of exposure. In the current method we monitor N-acetyl-S-(2-hydroxyethyl)-L-cysteine (HEMA), N-acetyl-S-(3-hydroxypropyl)-L-cysteine (HPMA),

S-(1-hydroxy-3-buten-2-yl)-N-Acetyl-L-cysteine (MHBMA), N-acetyl-S-(3,4-dihydroxybutyl)-L-cysteine (DHBMA), N-acetyl-S-(2-carboxyethyl)-L-cysteine (CEMA), and N-acetyl-S-(phenyl)-L-cysteine (PMA), N-Acetyl-S-(benzyl)-L-cysteine (BMA), 2-thioxothiazolidine-4-carboxylic acid (TTCA), N-Acetyl-S-(N-methylcarbamoyl)-L-cysteine (AMCC), N-Acetyl-S-(2-carbamoylethyl)-L-cysteine (AAMA) and N-Acetyl-S-(trichlorovinyl)-L-cysteine (TCVMA) in human urine. These analytes are metabolites of 1,3-butadiene (MHBMA, DHBMA), benzene (PMA), toluene (BMA), acrylamide (AAMA), carbon disulfide (TTCA), N,N-dimethylformamide (AMCC), acrolein (CEMA, HPMA), tetrachloroethylene (TCVMA), acrylonitrile, vinyl chloride, and ethylene oxide (HEMA). For matrix spike experiments the mean accuracy ranges from 98-107% and the mean percent difference ranges from 0.43-9.54%. The limit of detection ranges from 0.01-0.21 μg/L. By spiking urine with pure isomers and retention time interpretation, we could identify the correct diastereoisomer of MHBMA in human urine as S-(1-hydroxy-3-buten-2-yl)-N-Acetyl-L-cysteine. We applied this method to 690 urine samples collected (10 samples each) from 25 smokers and 44 non-smokers (categorized based on blood 2,5-dimethylfuran levels) to find that smokers have significantly elevated levels of AAMA, CEMA, DHBMA, HEMA, HPMA and PMA.

PS 1302 TOXICOGENOMIC IDENTIFICATION OF BIOMARKERS OF ACUTE RESPIRATORY EXPOSURE TO SENSITIZING AGENTS.

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Allergy induction requires multiple exposures to an agent. Therefore the development of high-throughput or in vitro assays for effective screening of potential sensitizers will require the identification of biomarkers. The goal of this preliminary study was to identify potential biomarkers that differentiate the response to allergen vs non-allergen agents following an acute exposure in naïve individuals. Female BALB/c mice received a single intratracheal aspiration exposure to Metarhizium anisopliae crude antigen (MACA) or bovine serum albumin (BSA) in Hank's Balanced Salt Solution (HBSS) or HBSS alone. Mice were sacrificed after 1, 3, 6, 12, 18 and 24h. Bronchoalveolar lavage fluid (BALF) was evaluated to determine total and differential cellularity, total protein concentration and LDH activity. RNA was isolated from lung tissue for microarray analysis and RT-PCR. MACA administration induced a rapid increase in BALF neutrophils, lymphocytes, eosinophils and total protein levels as compared to BSA or HBSS. Microarray analysis demonstrated differential expression of genes involved in cytokine production, signaling, inflammatory cell recruitment, adhesion and activation in 3h and 12h MACA-treated samples as compared to BSA or HBSS. Further statistical and pathway analyses allowed identification of ~100 candidate biomarker genes. Eleven genes were selected for further assessment by qRT-PCR. Of these, 6 demonstrated persistently increased expression (Ccl17, Ccl22, Ccl7, Cxcl10, Cxcl2, Saa1), while C3ar1 increased from 6-24h. In conclusion, a single respiratory exposure of mice to an allergenic mold extract induces an inflammatory response which is distinct in phenotype and gene expression from the response to a control protein. Validation of these biomarker genes with additional allergens and irritants is in progress. Biomarkers identified in these analyses will facilitate improvements in screening methods. (Supported by UNC/EPA Cooperative Training Agreement CR83323701. This abstract does not reflect EPA policy).

PS 1303 DEPLETION OF KUPFFER CELLS AS A MECHANISM FOR INCREASED SERUM ENZYMES.

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In clinical studies, evaluation of certain serum enzymes is routinely performed in order to assess the possibility of tissue injury. For example, an increase in the serum level of the enzyme alanine aminotransferase (ALT) is a sensitive indicator of hepatitis. However, changes in steady state levels of certain serum enzymes may reflect a change in either their rate of release to the bloodstream or in their clearance from the bloodstream. Since the turnover of many serum enzymes occurs via receptor mediated endocytosis by Kupffer cells (KCs) in the liver, it is possible that inhibition or depletion of KCs may also contribute to serum enzyme elevation. In order to better understand the role of KCs in serum enzyme clearance, KCs were depleted from rat liver using intra-venous injection of clodronate (CLO) liposomes. KC depletion was monitored using immunohistochemistry with antibodies to ED1 and ED2, which detect immature and mature (ED1) or just mature (ED2) KCs. ED2-positive cells were undetectable at 24 hours, with repopulation evident at 72 hours, and near to base-line levels by 8 days post CLO administration. The serum levels of ALT, aspartate aminotransferase (AST), creatine kinase (CK), glutamate dehydrogenase (GLDH), and lactate dehydrogenase (LDH) ALT were measured at 4, 8, 24, 48, 72, 96 hours, and 8 days post administration of CLO liposomes. The maximal increase was 8x at 8 hours for CK, and 4x, 10x, and 25x at 24 hours for AST, GLDH, and LDH, respectively with minimal changes in ALT. The increases in serum enzymes were inversely correlated to decreases in levels of KCs and returned to baseline by day 8. Histopathology of liver, heart, and skeletal muscle was normal and no changes to troponin 1 were noted, suggesting that CLO administration did not cause direct injury to these tissues. These data further demonstrate the role of KCs in serum enzyme clearance and support another mechanism for serum enzyme elevation that is not related to liver or muscle injury.

PS 1304 ANALYSIS OF LYMPHOCYTE SUBSETS IN PERIPHERAL BLOOD AMONG EXPOSED WORKERS AND PATIENTS WITH HYPERSENSITIVITY DERMATITIS INDUCED BY TRICHLOROETHYLENE.

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Trichloroethylene (TCE) is an important industrial chemical and is widely used. TCE is considered to have immune toxicity, hypersensitivity dermatitis have been described among workers exposed to TCE. The study consists of 16 patients with hypersensitivity dermatitis, 30 healthy TCE-exposed workers and 28 healthy workers unexposed to TCE. The lymphocyte subsets including CD4+ T cell, CD8+ T cell, B cell, NK cell, CD8+CD28+(-) T cell were measured in addition to the standard blood count analyses. All of the subjects in 3 groups were frequency matched by age and sex. The results showed that the absolute counts of lymphocyte, T cell, CD4+ T cell, CD8+ T cell, CD8+CD28- T cell were significantly increased in patients with hypersensitivity dermatitis compared with TCE exposed workers and unexposed workers, meanwhile, no significant differences in counts of lymphocyte, T cell, CD4+ T cell were demonstrated between exposed and unexposed groups. CD4+/CD8+ ratio and CD8+CD28+/ CD8+CD28- ratio were significantly decreased in both groups of hypersensitivity dermatitis and TCE exposed workers compared with unexposed group, both ratios were similarly in hypersensitivity dermatitis case and TCE exposed groups. The count of NK cell among 3 groups was in the increased tendency of unexposed control group >exposed group> case group, and the difference was significantly. No significant difference in count of B cell between 3 groups was found. These data provide evidence that occupational exposure of TCE causes change of lymphocyte subsets, especially T lymphocyte subsets. The counts of lymphocyte, T cell and CD4+ T cell might be biomarkers for screening cases with hypersensitivity dermatitis from TCE-exposed workers.

PS 1305 GROUP SPECIFIC COMPONENT: URINARY BIOMARKER OF SUBCLINICAL RENAL INJURY IN NEPHROTOXIN MODELS.

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Urine proteomics analysis by in-house efforts have identified Group Specific Component (GSC), also called Vitamin D binding protein, as a potential biomarker to early subclinical kidney degradation. To model subclinical renal toxin injury, low levels of D-serine were used to induce kidney damage at the proximal straight tubules. Male Fisher 344 rats were dosed intraperitoneally with control (0), 200, or 500 mg/kg D-serine in 0.9% saline and urine collected pre-dose and at timed increments post-dose. Renal damage was verified by histopathological examination of kidney tissue. Peptides based on rat GSC protein sequence were synthesized and used as antigens for polyclonal antibody development. Western blots using polyclonal GSC 1242 tested against urine from D-serine dosed rats demonstrated a strong signal as early as 12 hours postdose to a 52 kDal protein as well as a ~70 kDal protein. An examination of control urine demonstrated very low levels of the 52 and 70 kDal protein with background signal from secondary antibody alone negligible. GSC 1242 immunostaining of kidney tissue from animals dosed at 0 and 500 mg/kg D-serine confirmed that GSC protein is strongly induced in the kidney after dosing. To examine the GSC response to renal glomerular damage rather than the proximal tubule injury, urine samples from a rat study using puromycin were examined. The response of GSC in this nephrotoxin model significantly increases at 96 hours post-dose, and is seen at high levels up to 168 hours post-dose. Interesting, an examination of differential RNA expression of dosed versus control kidney demonstrated that GSC expression decreases upon nephrotoxin exposures. These pre-validation studies on rat group specific component indicate that it is present in the urine at low levels which dramatically increase upon exposure to both puromycin and D-serine, an indication that GSC response is not localized to proximal tubule damage.

was collected prior to dosing and at 12 hour intervals for a total of 168 hours. Urine samples were assayed by ELISA for clusterin, retinol binding protein (RBP) 4, heme oxygenase (HO)-1, osteopontin (OPN), Yb1 (mu) glutathione S-transferase (GST), α glutathione S-transferase (αGST), metalloproteinase tissue inhibitor (TIMP)-1 and β_2-microglobulin. Neutrophil gelatinase-associated lipocalin (NGAL) and kidney injury molecule-1 (Kim-1) were assayed by Meso Scale Discovery (MSD) Multi-Spot® Assay. Biomarker levels remained constant for control animals throughout the time course. However, for animals dosed with 200 and 500 mg/kg D-serine, significant increases were observed with peaks at 12 hours post-dose (HO-1, Yb1 GST and αGST), 24 hours post-dose (clusterin, RBP4, TIMP-1 and β2-microglobulin), 96 hours post-dose (Kim-1 and NGAL) or 120 hours post-dose (OPN). Biomarkers returned to baseline levels at 36 hours (Yb1 GST and αGST), ≥ 48 hours (HO-1, β_2-microglobulin, TIMP-1, clusterin and RBP4) or ≥ 168 hours (NGAL, Kim-1 and OPN). Gene expression studies were also conducted in control and dosed kidney tissue, and significant increases in transcription were seen in most biomarkers examined. RBP4, however, demonstrated significantly lower expression upon nephrotoxin exposure. Expression profiles indicate that this protein set differed in maximal response times. Their collective detection in urine is a potential noninvasive strategy to determine early onset of low level subclinical kidney damage in response to toxin exposures, ultimately leading to development of rapid field monitoring for the prediction of health hazards associated with chemical exposure.

PS 1879 TRANSPLACENTAL DISTRIBUTION OF METALS AND THEIR INTERACTIONS ASSESSED BY BIOMONITORING IN MOTHER/CHILD PAIRS.

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Exposure of the fetus to (heavy) metals has been associated with adverse health outcomes including developmental toxicity. However, few data exist on the transplacental passage of metals and their interaction with each other in the maternal-fetal unit. In our study, venous and umbilical cord blood samples from 50 mother/child pairs were studied for exposure to multiple heavy metals, essential minerals and trace elements. Smoking status was assessed by cotinine in urine. Lead (Pb) showed the highest median concentration of heavy metals in maternal samples (11.5 µg/L) followed by nearly equal concentrations of mercury (Hg, 0.44 µg/L) and cadmium (Cd, 0.34 µg/L). Smokers showed higher Cd levels than non-smokes (0.73 vs. 0.29 µg/L, P<0.001). Slightly but significantly lower levels of Pb were observed in fetal blood (10.3 µg/L, P<0.004), whereas Cd was strongly reduced (0.05 µg/L). In contrast, higher concentrations of Hg were detected in fetal samples (1.48 µg/L, P<0.0001). Selenium (Se) and iron (Fe) showed a similar distribution in the maternal/fetal unit as observed for Pb, whereas the distribution of manganese (Mn) was similar to Hg. Copper (Cu) and Zn were strongly reduced in the fetus and distribution was more similar to Cd. Linear regression analysis revealed positive associations between maternal and fetal concentrations for Pb, Mn and Hg (P≤0.014). No associations between maternal and fetal blood were found for Cd, Cu, Fe and Zn. Exposure to heavy metals (single or in combination) did not influence the levels of essential minerals such as Zn. In conclusion, the placenta provides a barrier for Cd, Cu and Zn, whereas Fe, Pb and Se enter the fetal environment unaffected. Mn and Hg are unequivocally transported to the fetus resulting in increased exposures compared to the mother. However, homeostasis of essential elements remains unaffected by exposure to heavy metals at low exposures. Overall, our results contribute to the risk assessment of heavy metals and adverse health outcome in the most vulnerable population, the fetus.

PS 1880 SURVEILLANCE FOR SYSTEMIC EFFECTS OF METALS AND OTHER MATERIALS RELEASED FROM RETAINED EMBEDDED FRAGMENTS IN U.S. SOLDIERS.

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Concern has heightened regarding long term health effects associated with embedded fragments in soldiers. In the past, fragments embedded in muscle tissue were thought to be relatively inert, however recent work has shown that veterans with embedded depleted uranium (DU) fragments have elevated blood and urine uranium levels more than 18 years after injury involving DU munitions during the first Gulf War. This finding is supported by studies showing release of metals from certain types of medical implants. To better understand and prevent health problems resulting from retained metal and non-metal fragments in soldiers, the Department of Veterans Affairs has established a program charged with developing clinical management guidelines for embedded fragments. These will be based on results from analysis of fragment content, health surveillance and biomonitoring of veterans with prolonged systemic exposure to chemicals released from fragment material over time. Chemical characterization of over 400 removed fragments has shown that most are metal alloys (83%) while others are different types of organic material, plastics, wood and stones. Based on this information and knowledge of the toxicity of metals, a biomonitoring protocol utilizing primarily urine has been developed to characterize systemic exposure to the following carcinogenic and cytotoxic metals: Al, As, Cd, Cr, Co, Cu, Fe, Mn, Ni, Pb, U, W and Zn. Customized health surveillance and management guidelines will be developed for veterans with chronically elevated excretion of specific metals using biomarkers of potential effects of the metal(s) of concern. Biomonitoring protocols for compounds released from non-metallic fragment materials, such as isocyanate, phthalates and acrylics, will continue to be developed as our knowledge of the breakdown of fragments embedded in muscle tissue increases. *Supported by Department of Veterans Affairs and the Armed Forces Institute of Pathology*

PS 1881 DOD IMPACT ASSESSMENT AND MANAGEMENT OF NAPHTHALENE-RELATED RISKS.

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The Department of Defense Chemical and Materials Risk Management Directorate is using a scan-watch-action process to identify, rank and manage risks associated with emerging contaminants. Naphthalene is characterized as a likely human carcinogen by the NTP and in the EPA's most recent draft health risk assessment. Thus, naphthalene-related environmental health regulations are evolving. The potential impacts have been assessed, using multi-criteria decision analysis, for five of the Department's functional areas. One of the areas of concern is exposure to naphthalene among fuel handlers. To determine whether these exposures present unacceptable risk, the Army Research Office awarded a Small Business Innovative Research Project for the development of a miniature real-time naphthalene sensor. NIOSH's Biomoniting Team and Investigators from the Army Research Institute for Environmental Medicine, UC-Davis and the Army Corps of Engineers are collaborating on a second DOD-funded project. This project will validate the prototype sensor as a dosimeter by defining correlations between measured exposures and biomarkers of exposures to-be-collected from military fuel handlers. To date, naphthalene specificity with sensitivity of 0.5 mg/m3 has been demonstrated and definition of the firmware chemometrics is underway. Implementation of the human subjects research protocol is pending institutional review boards' approval.

PS 1882 EFFECTS OF STYRENE CO-EXPOSURE ON FORMATION OF 1, 3-BUTADIENE DERIVED N7-GUANINE ADDUCTS.

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Protein and DNA adducts have been widely applied for monitoring the internal dose of reactive compounds and metabolites after environmental or occupational exposures. Formation of DNA and protein adducts correlate well with external exposures in rodents and human studies. A recent study in butadiene (BD) exposed workers demonstrated that BD-specific protein adducts correlate with external BD exposure (R2 = 0.6) in BD monomer workers and not in BD-styrene polymer workers (R2= 0.08), despite the fact that the BD exposures were 3-fold higher in the polymer workers. Styrene co-exposure was 14-fold higher in the polymer workers than in the monomer workers. It is suggested that styrene co-exposure effects BD metabolism, since both are metabolized by P450 2e1 to DNA reactive epoxides. Subsequent in vitro studies showed inhibition of P450 2e1 activity by styrene oxide. We report herein the effects of styrene co-exposures on the formation of N7-guanine adducts in vivo. Female B6C3F1 mice were exposed to filtered air, 20 ppm BD, 250 ppm styrene or 20 ppm BD plus 250 ppm styrene for 6 h /day, 5 days/week for 2 weeks. A method was developed for simultaneous quantitation of the isomeric N7-hydroxybuten-guanine (N7-HB-Gua), N7-trihydroxybutan-gua-